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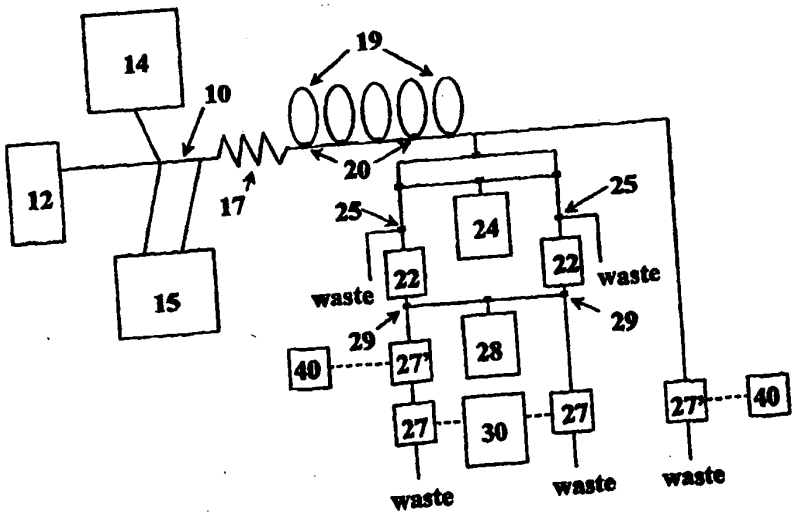

(57) Abstract

The present invention relates to an assay apparatus for detecting substances in a liquid carrier. The apparatus comprises a sample input means and a plurality of primary detector means located in a flowpath. The plurality of primary detector means consists of at least two different detectors selected from the group consisting of: a) fluorescent detectors; b) UV spectrometers; c) infra–red spectrometers; d) Raman spectrometers; e) other spectrometric detectors; f) electrolyte determination detectors; and g) other electro–chemical detectors. Each primary detector means is located in a separate detector pathway of the flowpath.

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An Assay Apparatus with Multiple Detectors

The present invention relates to an assay apparatus for determining whether or not a particular substance is present in a sample. In particular the present invention relates to an assay apparatus where certain substances are tested for in a liquid carrier, such as in a flow injection assay system.

A great number of different techniques exist for performing assays and the instances in which assay techniques are used are becoming more widespread. An overview of currently used techniques is given in the article "Update on immunoassay automation" Proc. UK NEQAS Meeting 1994:1:163-170; Wheeler, Michael J. For example, various types of immunoassays are now commonly used to test blood and other samples for a great number of different compounds. Yet, even a large hospital may not have a large number of different assay machines and so the apparatus must be capable of performing a range of different tests on different samples. The versatility of any assay technique must necessarily increase as the number of tests that can be requested on samples increases. The growing desire that assays can be performed directly in places such as Doctors Surgery's, rather than sending the samples away for analysis by a laboratory in a hospital further increases the demand for versatile machinery.

In conjunction with the growth of the assay systems, it has become necessary that any particular assay is performed ever more precisely. The importance of the accuracy of the test will be apparent, as, for example, a patient's treatment may be determined based on the result of the assay and so an inaccurate result may lead to inappropriate

treatment of the patient.

The growth in assay techniques being used has led to a large number of systems on the market. Presently, most systems are semi-automated or automated systems, where after loading of the reagents and samples no further input is required from a human operator, unless a breakdown occurs.

As will be apparent from the aforementioned article by Wheeler, the majority of the devices presently on the market comprise a loading tray for loading multiple samples, for example between 20 and 100 samples, which are not necessarily of the same nature or having the same assay performed on them. There is also a reagent input tray which holds a number of reagent cartridges for the various different tests to be performed. In the machine the samples are transferred, normally by pipetting into an assay cell where the sample is combined with the necessary reagent or reagents. The assay cell is then transferred to a part of a machine where it can be held for sufficient time for the reagent and the sample to combine. Thereafter the sample cell is transferred to the detector which detects the presence of a known indicator to determine whether or not the sample contained a particular component and/or how much of that component was present in the assay. Normally a robotic arm is used for transferring the assay cell around the machine, for example from the loading area to the wash station, to the waiting and onward to the detector. Whilst assay apparatus of this type can offer semi-automated functioning, problems do occur due to the mechanical movement of the samples. Furthermore, it is necessary to have pipettes with replaceable pipette tips or other means to ensure that one sample does not contaminate

another sample when being analysed. As each sample cell must be incubated with the appropriate reagents, a relatively large number of sample cells may have to be incubated at any one time and thus the size of the machine remains relatively large due to the space required for the waiting area.

It is well known to use fluorescence spectroscopy and microscopy to determine the concentration of analytes in a sample by measuring the emission signal for changes in intensity or wavelength shifts. Various methods and apparatus are known for this purpose such as those described in US Patent Nos. 4,877,965 and 5,196,709.

Fluorescence detection is particularly useful in biological tests where sample size is often very small, e.g. in assaying for nucleic acids or proteins. In these circumstances it is very important to minimise interference from background radiation and techniques have been devised as described, for example, in US Patent Nos. 4,006,360, 4,341,957 and 4,791,310.

Recently, it has been suggested to use flow injection technology in assay equipment. A review of this technology is given in an article by Puchades, R. et al "A Comprehensive Overview on the Application of Flow Injection Techniques in Immunoanalysis, Critical Reviews in Analytical Chemistry" 23,(4):301-321(1992). The fundamental difference between this technology and the above described systems is that the samples and the reagents are combined in a fluid stream, which extends to the detector. This dispenses with the need of sample cells and generally reduces the mechanical components of the system.

Th present invention seeks to provide an assay apparatus which allows versatile measurement of analytes in a sample.

According to the present invention there is provided an assay apparatus for detecting substances in a liquid carrier, comprising a sample input means and a plurality of primary detector means located in a flowpath, wherein the plurality of primary detector means consists of at least two different detectors selected from the group consisting of: a) fluorescent detectors; b) UV spectrometers; c) infra-red spectrometers; d) Raman spectrometers; e) other spectrometric detectors; f) electrolyte determination detectors; and g) other electro-chemical detectors, and the primary detector means are located in separate detector pathways of the liquid flowpath. The apparatus thus provides a much larger number of tests to be performed than conventional apparatus whilst still being housed in a single unit. Many components can be shared upstream of the detectors, in particular in the control and input side of the apparatus.

The various detectors are selected from one or more of the detectors utilising spectrophotometric and/or electrochemical principles. In these circumstances it is possible that the analysis may be more simple than immunoassays relying on fluorescence detection and so the use of immobilised reagents and the barrier separation device can be avoided, but normally barrier separation will be provided to allow immunoassay testing as described in International application No. PCT/GB97/00334, the contents of which are hereby incorporated by reference.

In order to achieve more efficiency and control in the apparatus the flowpath may include incubation means

preferably downstream from mixing means provided for mixing the sample with desired reagents. Incubation can, however, take place in the main flowpath provided that the sample and reagent have sufficient time to interact prior to the selected detector as primary detector means.

The sample can preferably be split so that different types of detection can be carried out on a single elution peak. Additionally, a secondary detector can be located in a single detection pathway. It may be appropriate to carry out analysis on a single sample where different types of test are applied together with at least two different detectors measuring analytical signals from the target analytes in the sample and under these conditions it can be beneficial to incorporate the additional secondary detectors into a flowpath leading to e.g. a fluorescence detector.

It is preferred if the apparatus provides for multiple analyte detection of a single sample.

The fluid pathway is preferably divided into a plurality of the said detection pathways with splitter-valve means arranged to direct each respective aliquot into a respective selected one of the detection pathways. An aliquot may be formed from a whole or part of a sample. The multiple pathways may extend from any point after the incubation loops or often if included in the particular apparatus. Multiple pathways improve the capacity of the apparatus as one pathway can be washed whilst another is analysing a sample.

Advantageously, one detector comprises the detection means for several, e.g. all, of the detection pathways where the

same detection principle is employed.

Many chemical tests rely on the principle of absorbance as the analytical measurement where for example a colour change occurs in a reagent in the presence of the target analyte. One variation where this approach can be applied is in the determination of endogenous enzyme or other protein levels with absorbence measurements of a colorimetric substrate indicating the relative amount of enzyme or protein in the sample. In the flow system described hereinafter the analysis could be carried out by holding the reagent/sample mixture in the incubation loops to allow for the generation of the coloured product. After this time the mixture would be released into the flow stream and directed to the flow path that leads to the spectrophotometer where the absorbence measurement is made.

In addition to measuring endogenous proteins there is often a requirement to determine the levels of electrolytes in blood samples and this may be carried out through the use of ion selective electrodes. The operation of these devices is well known and their incorporation into the flow stream at any point would be a simple matter with electrolyte measurements made as the sample passes over the electrode. Often such electrodes will form secondary detectors in the apparatus.

Preferably at least some of the detection pathways include the incubation means as in this way the different reagent will not contaminate separate detection pathways. Advantageously the detection pathway begins before the reagent is added to the liquid in the flowpath in at least some of the detection pathways. This is advantageous where the reagents interact with one another as it enables a

single sample to be split into several aliquots and tested with several reagents which would otherwise lead to inaccuracies in the multiple analysis of the sample.

Preferably the detectors are solid state detectors. The small size of these detectors advantageously allows the apparatus to be contained in a single housing. Laser diodes are suitable for applications such as fluorimeters. Light emitting diodes may be suitable for most spectrochemical applications.

Preferred embodiments of the present invention will now be described with reference to the accompanying drawings, in which:

Fig. 1: is a schematic representation of an assay apparatus according to the present invention;

Fig. 2: illustrates a preferred embodiment of a detector for use in the apparatus of Fig. 1;

Fig. 3: depicts the overall construction of a flow injection apparatus of the preferred embodiment of the present invention.

Figure 1 shows a preferred embodiment of the present invention comprising a flow injection immunoassay analyser as an integrated system. The system operates using the principles of flow injection analysis, that is a continuous stream of liquid is used to transport discrete volumes of sample or reagents that are injected into the stream. These materials can then be brought into contact with one another or with other materials that may be in solution or fixed to a surface so that they interact in a way that can be measured and thus the flow injection process is directly analogous to the manipulations that take place in traditional immunoassays using microtiter plates or tubes

except that injection loops or syringes and precise control of flow rate replace the use of pipettes, washers and shakers.

A carrier buffer stream is generated from run buffer 12. The carrier buffer should not contain any gas as this leads to inaccuracies in the instruments. A plurality of samples are held in a sample processing unit 14 which also prepares each sample for analysis. Analysis for a particular target molecule (a product) takes place by injecting a known volume of a sample that possibly contains the product, into the carrier buffer stream and mixing it with reagents from a reagent cartridge 15. There may be several different inlets for the reagents. Some of these inlets may be in particular detection pathways. The inlets may be associated with more than one reagent source.

The sample processor unit 14 has the capacity to hold approximately 100 samples and a normal variety of tube sizes. The unit 14 is capable of carrying out accurate and precise pipetting to generate a sample dilution as required. This may be in a traditional manner with appropriate volumes transferred to a separate tube on the processor bed or by using the flow system where a fixed volume of sample and a variable volume of diluent (or vice versa) are merged in a mixing coil before a fixed volume is taken for analysis. The unit 14 may employ a robot arm (not shown) carrying a sample probe (not shown). The robot arm would normally be capable of movement in three planes and the probe can be washed between sample manipulations at an on board wash station. Samples can be loaded onto the processor unit 14, preferably in their original tubes, of varying dimensions, in pre-prepared racks of tubes or in pre-prepared microtiter plates. Sample identification and

tracking is made possible through bar codes which may be placed on the individual tubes, or the tube racks or the microtiter plates and the bar codes are read by the on board bar code reader, though other tracking systems can be used.

The flow system of the instrument consists of transmission tubing 10 made from chemically and biologically inert material such as commercially available nylon or PEEK with an inner diameter of typically 0.8mm, although this may change to suit the circumstances. The pumping system (not shown) consists of several low pressure pumps, most likely peristaltic pumps which may be of differing size, sophistication and performance and will be capable of delivering a highly reliable flow rate. A central pump will be used to move the carrier buffer, samples and reagents through the system whilst other pumps which are likely to be less sophisticated, will be used to carry out other manipulations, such as reagent transport, barrier washing and conjugate elution. The operation of each pump will be controlled by the central computer (not shown) to ensure optimum performance and effective synchronisation. The computer will also have control over the many automated switching valves (described in more detail hereinafter), which at the appropriate times direct samples or reagents into or out of the main carrier stream. These valves may be electronically or pneumatically operated and must be extremely reliable and robust as they will be used many times in any working day. They will be very simple in design needing only to switch the liquid flow between one of two channels or limited number of channels. The valves must have chemically and biologically inert surfaces where they come into contact with the liquid stream.

Generally, the reagents required for each assay are specific for the analyte of interest, however the same principles are applied in each case and only two components are normally required.

If the assay is based on fluorescence detection, it is preferred to utilise microbeads of a defined diameter and with the property of neutral density so that they remain in suspension and it is likely that the beads will be made of a cellulose material with low non-specific binding properties although other suitable materials may be preferred. The surface of these beads is coated, probably through covalent conjugation although other procedures such as adsorption may be possible, with a ligand binder material such as an antibody or other compound that specifically binds only the analyte of interest in the assay. In a more sophisticated assay format, it is possible to coat two or more ligand binders with different specificity's onto the bead surface and this allows for multianalyte determination from the same sample. The second reagent is a labelled material which may be an analogue of the analyte of interest or binder with specificity for the analyte depending on the assay format required. The label is often a fluorophore with spectral characteristics that allow it to be detected in the near infra red region of the electro-magnetic spectrum, however other labels such as liposomes, enzymes and chemiluminescent materials are also possible.

The fixed volumes of the reagents and the sample are mixed together in a mixing coil 17 and allowed to incubate together for a fixed time. If microbeads are used in the assay format, these beads will be included in the incubation process.

The incubation can take place just in the main flow, for example when the incubation time is short. The incubation is preferably accomplished by removing the aliquot out of the main flow stream and into one of the incubation loops 19. The access to the incubation loops 19 is controlled by valve means 20. The loop 19 is made from a fixed length of transmission tubing of an appropriate internal diameter, however the overall volume should be carefully chosen to ensure precise replication of incubation conditions. During incubation any of the product which is contained in the sample should interact with the reagent to form a complex. The complex will be bound to said microbeads if these are used in the particular assay. The complex must include a detectable moiety. In some cases a particular incubation loop may form part of a detection pathway and is used only for incubation of mixtures for the particular detection pathway.

After time for incubation, the computer allows the aliquot of mixture to be carried down to the detectors.

In assays using microbeads on which the complex is formed, a membrane barrier 22 is used to separate the complex from the sample. The membrane barrier 22 retains the microbeads whilst all other materials flow through and are passed to waste. The barrier 22 consists of a porous membrane made from a chemically and biologically inert material such as nylon and the barrier 22 is sized and arranged to prevent all beads from passing through to the flow cell. The pore structure of the membrane is governed by the size of the microbeads, however it is important that the membrane has a low non-specific binding of excess reagents and that substantially all (eg>95%) of the beads are retained. The flow path is then washed by a period of flow with carrier

buffer to wash off any unbound reagents and possibly the flow rate is raised during this period, so the barrier 22 should have good flow properties.

Subsequently, the various valves are switched in synchronisation to divert the main buffer flow from the barrier 22 whilst introducing an elution buffer from vessel 24 to flow through the barrier 22. The switching of the valve can be gauged from monitoring the unbound reagent flowing through the flow cell 27 to waste. This releases the label (detectable moiety) from the microbead to flow through the barrier 22. The flow is now directed to a flow cell 27 for measurement downstream. Following elution further valve switching allows the membrane to be back flushed with an appropriate buffer from vessel 28 which removes the beads to waste through valve 25 and cleans the membrane ready for the next sample aliquot.

After incubation the sample mixture will be carried to a detector 30,40 chosen from: a) fluorescent detectors; b) UV spectrometers; c) infra-red spectrometers; d) raman spectrometers; e) other spectrometric detectors; f) electrolyte determination detectors; g) other electrochemical detectors. The apparatus will have at least two different types of detector. It is preferred if the detectors are solid-state detectors in view of the compact nature of these devices. For example, the microspectrometer available from MicroParts described in "Opto and Laser Europe", pages 70,71, Issue 41, June 1997 could be incorporated. Also, ion-selective electrodes may be used for example as described in "Use of ion-selective electrodes as detectors in flow-injection analysis", Magalhaes e Adelio, J.M. et al., Port. Electrochim. Acta, Sep 1991, Vol. 9, pages 429-467.

If microbeads are not used in the particular assay format, then the particular aliquot can be directed to the detector at any stage provided that sufficient time is allowed for the reagents and sample to interact. For example, in the case of ion selective electrodes being used as one type of detector, then this detector 40 could be located anywhere downstream of the input as no reagents are required for this detector technique. If a particular detector is not associated with a particular detection pathway, then the detector will normally form a secondary detector. Another type of secondary detectors are those detectors which are placed in a particular detection pathway, but is not the primary detector in said pathway. This can occur when the secondary detector and associated reagents (if any) do not interfere with the primary detector and are not affected by the reagents etc. incorporated for the primary detector.

It is preferred if capability is given in the apparatus for microbead separation for some assay formats. In that case it is then convenient if valve means are provided to direct a particular aliquot to a predetermined detector 40 downstream of the barrier 22. There are thus plural limbs of the flowpath downstream of the barrier.

The detectors 30/40 may require different types of test cell. For example a fluorimeter test cell would normally comprises a quartz silica cylinder, although other materials and shapes may be preferred, with a total volume unlikely to exceed 200μl and which is normally illuminated by a light source and monitored by a detector 30 as explained in more detail below.

In most cases each limb (detection pathway) of the flowpath will contain one test cell, but a plurality of different

test cells can be provided on a single limb, for example if a histogram of analyte determination is required. Different types of detector may be located on the same or separate limbs.

A particular preferred detector is the detector 30 which comprises a fluorimeter shown in greater detail in Fig. 2.

The detector 30 is shown in greater detail in Figure 2 and consists of a laser diode module 31, 32, 33, a light path of mirrors and beam splitters 38 to 42, identical duplicate flow cells 27 and a single sensor 35 as described hereinbefore. Although two flow cells 27 are shown, an instrument employing the detector 30 may include one, two, three or more flow cells depending upon the capacity required for the machine. If there is only one flow cell then the valving upstream of the detector can be simplified as can the controlling software which can reduce the cost of the apparatus for situations where only a low capacity apparatus is required. On the other hand a greater number of flow cells may increase the capacity of the apparatus, but additional flow paths through the apparatus may be required to fully exploit the greater detector capacity. The detector 30 will often be based when a membrane barrier is used in the assay system.

In the illustrated embodiment there are two detector flow pathways to detectors 30. This is particularly advantageous as one of the two pathways can be analysing a sample whilst the other pathway is being washed from wash buffer 28 via one of the wash valves 29. This greatly increases the number of samples which can be analysed in a given time period. This feature is also true when different detection principles are utilised in the

different detection flowpaths. This design is particularly advantageous when used in conjunction with the detector 30 of the present invention which allows for the two (or more) flow cells 27 to be analysed from a single radiation generator/emitter and so the increased capacity is provided at little extra cost. This feature of multiple test cells for a single detector can also be applied to the other detectors 40.

The choice of lasers will be very much dependent on the available fluorophores since the lasing wavelength and optimum fluorophore excitation wavelength need to be well matched. The rate of development in the field of solid state lasers and appropriate fluorophores is rapid and the final choice for these components cannot be made now. However it is likely that the lasers should have at least a 1 milliwatt output (preferable 10 milliwatt) and operate above 400nm, whilst the fluorophores should be water soluble if an aqueous carrier buffer is used, stable in solution, unaffected by pH changes, emit their fluorescence above 600nm and have the general properties required of a good fluorophore. Various fluorophores are known in the art and more are being developed. For example see the review article by Fabian, J., et al, Chemical Reviews, 1992, 92, 1197-1226 which gives details of a number of different types of available fluorophores with some spectral details. Similarly, various suitable light sources are known (i.e. laser diode modules) or are being developed. The skilled person can pair light sources with respective fluorophores depending upon the particular requirements for the pair required at the time.

The laser module 31, 32, 33 can contain more than one laser, each of which can in turn be switched into the light

path whilst at the same time collecting data from the detector into a separate channel. Computer control of this switching allows the potential for multi-label detection by operating 2 or more carefully chosen lasers of different excitation wavelength in a rapid pulse mode, e.g. one after the other, and monitoring the associated emission from its paired fluorophore. In this way specific measurements can be made in mixtures of fluorophores and this leads to the possibility of multi analyte determinations from the same elution peak. If pairs or more of analytes are measured in this way the throughput of the instrument is greatly increased and the usage of sample greatly reduced since mixed specificity beads can be used for the sample capture. The resulting signal is plotted as a peak and the calculated area used to determine the concentration of the sample from a curve generated from standard solutions.

For further details of the preferred fluorimeter please see the applicants' co-pending application filed the same day as the present application and entitled "A detector", the contents of which are hereby incorporated by reference.

The system operates in random access mode but has the in-built capacity for immediate analysis of emergency samples, which are placed in separate racks on the autosampler. The timing and scheduling of operations are precisely controlled by the software which is icon driven, intuitive to use and which operates in a Windows™ environment. The software is designed to run on a notebook type computer which can be closed and stored in the base of the instrument when not required. Communication with the instrument is bi-directional, allowing feedback from off-scale results to initiate appropriate dilution and sample re-analysis. The instrument and software are fully

configured for operation within a Laboratory Information Management System (LIMS) environment, including quality control monitoring of assay controls and reagent cartridge performance.

In many cases, a single sample will be carried to a single detector for a single analysis. However, it is preferred if some or all of the detectors allow multiple determination on a single sample by detecting more than one detectable moiety in a single elution peak. Additionally or alternatively the single elution peak containing a single sample may be split between different detectors for different types of analysis. These different detectors can be contained in the same or different detector pathways of the flowpath.

It is preferred if the assay apparatus comprises a flow injection system. Yet the invention can be implemented by just providing a flowpath consisting of the various detector pathways. The sample is the input directly into the selected pathway or pathways, if the sample is split. In this case it would be normal for incubation with any reagent to occur prior to input to the assay apparatus, but incubation can take place in the limb if the sample is held in the limb for sufficient time. In this embodiment mixing of the reagents and sample can take place separately, e.g. in cuvette.

The analyser is designed to be capable of measuring greater than 20 clinically important substances, each of which will have a dedicated cartridge of reagents, capable of approximately 200 analyses held in the reagent carousel on board the instrument. The cartridge design will ensure that reagents can be stirred if required and kept at

constant temperature through control of either the carousel compartment or the cartridge itself. Each cartridge holds information about itself, possibly on a bar code.

Claims:

1. An assay apparatus for detecting substances in a liquid carrier, comprising a sample input means and a plurality of primary detector means located in a flowpath, wherein the plurality of primary detector means consists of at least two different detectors selected from the group consisting of: a) fluorescent detectors; b) UV spectrometers; c) infra-red spectrometers; d) Raman spectrometers; e) other spectrometric detectors; f) electrolyte determination detectors; and g) other electrochemical detectors and each primary detector means is located in a separate detector pathway of the flowpath.

2. The assay apparatus according to claim 1, wherein the flowpath extends from the input means via incubation means to the detector means.

3. The assay apparatus according to claim 2, wherein the incubation means comprises incubation loops which comprise a said flowpath into which part of the fluid in the main flowpath (e.g. a chosen discrete aliquot) is flowed for a predetermined time.

4. The assay apparatus according to claim 2 or claim 3, wherein the incubation means forms at least part of the direct flowpath between the input means and the detector means.

5. The assay apparatus according to any one of the preceding claims further including mixing means for mixing the sample with desired reagents.

6. The assay apparatus according to any one of the

preceding claims, wherein the input means is arranged to input test samples in discrete aliquots into the flowpath, and the apparatus further including valve means arranged to selectively direct part or all of each discrete aliquot to a predetermined primary detector means.

7. The assay apparatus according to any one of the preceding claims, wherein at least one of the primary detector means comprises a fluorimeter.

8. The assay apparatus according to any one of the preceding claims, wherein at least one detector is arranged to perform multi-analyte determination of a sample.

9. The assay apparatus according to claim 7 or claim 8, wherein the flowpath upstream of the detector includes barrier means to physically prevent flow of particles of greater than a particular diameter.

10. The assay apparatus according to any one of the preceding claims, wherein the apparatus is a flow injection assay apparatus.

11. An assay apparatus as hereinbefore defined with reference to and/or as illustrated in the accompanying drawings.

AMENDED CLAIMS

[received by the International Bureau on 12 January 1999 (12.01.99);
original claim 1 amended; remaining claims unchanged (1 page)]

1. An assay apparatus for detecting substances in a liquid carrier, comprising a sample input means for inputting sample into the liquid carrier and a plurality of primary detector means located in a flowpath, wherein the plurality of primary detector means consists of at least two different detectors selected from the group consisting of: a) fluorescent detectors; b) UV spectrometers; c) infra-red spectrometers; d) Raman spectrometers; e) other spectrometric detectors; f) electrolyte determination detectors; and g) other electro-chemical detectors and each primary detector means is located in a separate detector pathway of the flowpath and the sample is flowed to the or each primary detector in the said liquid carrier.

2. The assay apparatus according to claim 1, wherein the flowpath extends from the input means via incubation means to the detector means.

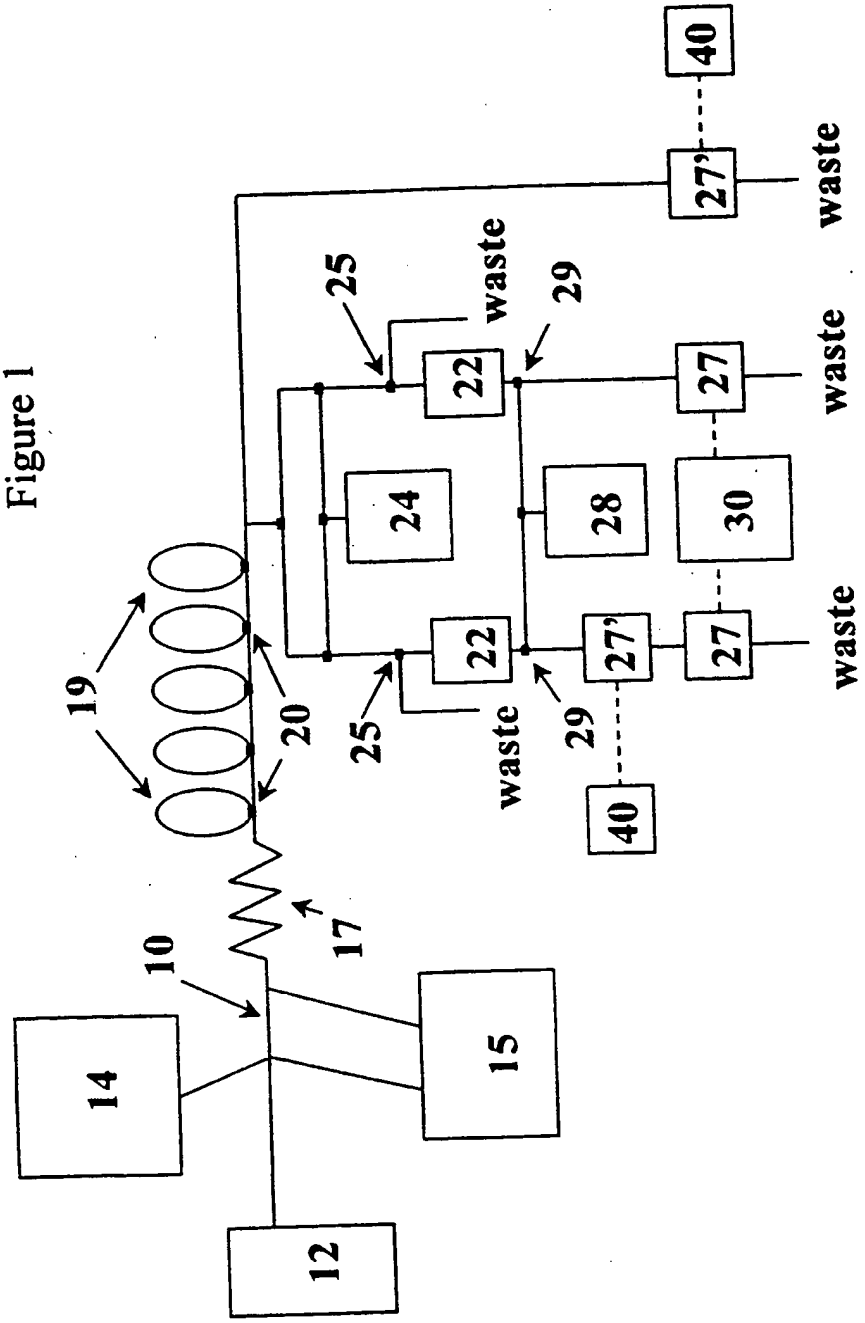
3. The assay apparatus according to claim 2, wherein the incubation means comprises incubation loops which comprise a said flowpath into which part of the fluid in the main flowpath (e.g. a chosen discrete aliquot) is flowed for a predetermined time.

4. The assay apparatus according to claim 2 or claim 3, wherein the incubation means forms at least part of the direct flowpath between the input means and the detector means.

5. The assay apparatus according to any one of the preceding claims further including mixing means for mixing the sample with desired reagents.

6. The assay apparatus according to any one of the

Figure 1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25
waste
29
40
17
10
14
15
12

Figure 2

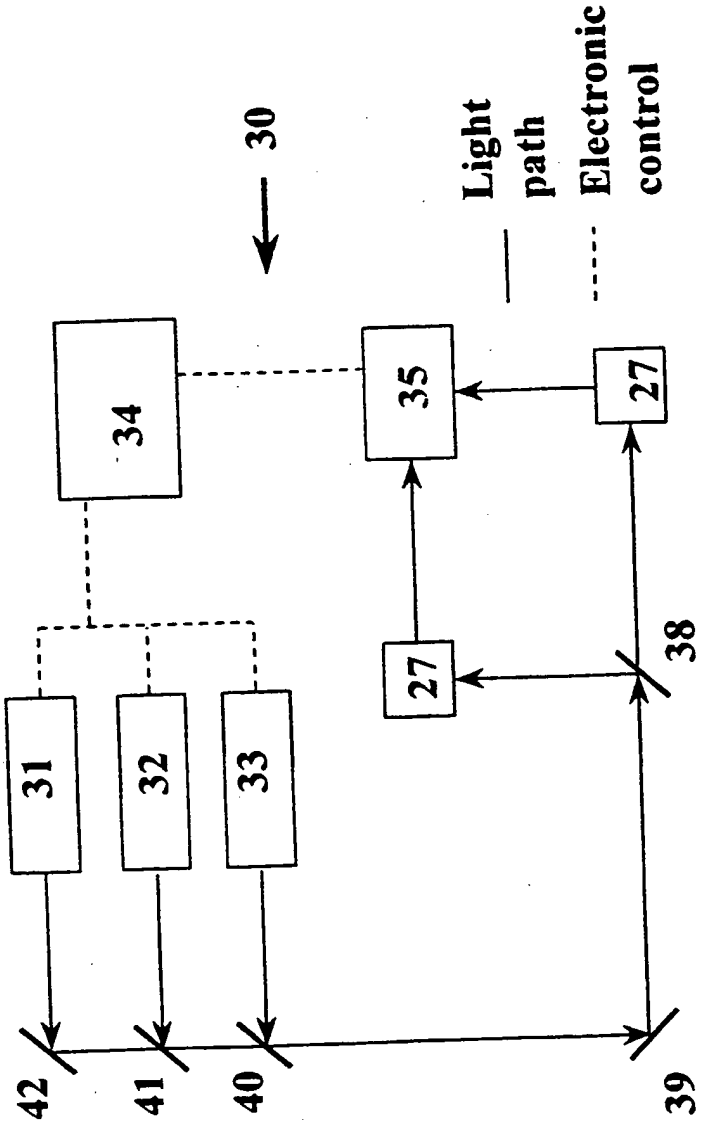
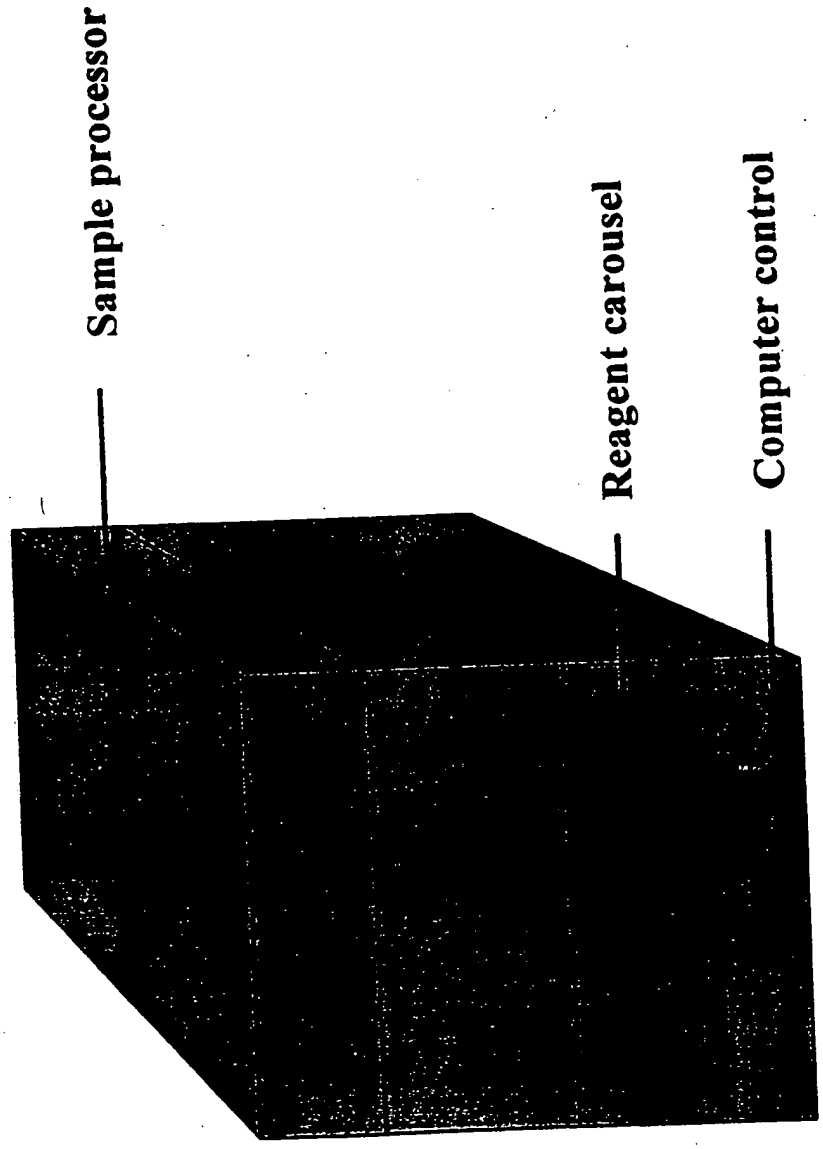

30
Light path
Electronic control
34
35
27
27
38
31
32
33
42
41
40
39

Figure 3



INTERNATIONAL SEARCH REPORT

Int ional Application No
PCT/GB 98/02396

A. CLASSIFICATION OF SUBJECT MATTER
IPC 6 G01N35/08

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 6 G01N

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	EP 0 375 153 A (FORD MOTOR CO ;FORD FRANCE (FR); FORD WERKE AG (DE); FORD MOTOR CO) 27 June 1990	1,5-7,10
Y	see abstract see column 3, line 14 - line 22 see column 4, line 47 - column 5, line 3 see column 7, line 43 - column 8, line 6 see column 8, line 40 - line 56 see column 9, line 21 - line 23 see figure 1	2-4,9
X	US 5 108 928 A (MENARD KEVIN P ET AL) 28 April 1992 see abstract see column 3, line 21 - line 65 see figure 1	1,6,8,10

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[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

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Date of the actual completion of the international search	Date of mailing of the international search report
4 November 1998	12/11/1998

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	Verdoodt, E

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INTERNATIONAL SEARCH REPORT

Int ional Application No
PCT/GB 98/02396

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

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Y	US 4 158 630 A (STEARNS STANLEY D) 19 June 1979 see column 2, line 25 - line 40 see figure 1	2-4
Y	US 5 372 783 A (LACKIE STEVE J) 13 December 1994 see abstract see column 5, line 64 - column 6, line 15 see column 5, line 49 - line 60 see figures 6,7	9
A	US 4 456 689 A (WITTY THOMAS R ET AL) 26 June 1984 see column 4, line 24 - line 50 see figures 1,2	2-4
A	US 4 272 483 A (SCHICK KARL G) 9 June 1981 see abstract see column 3, line 63 - last line see column 4, line 27 - line 59 see figure 1	2-4

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INTERNATIONAL SEARCH REPORT

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